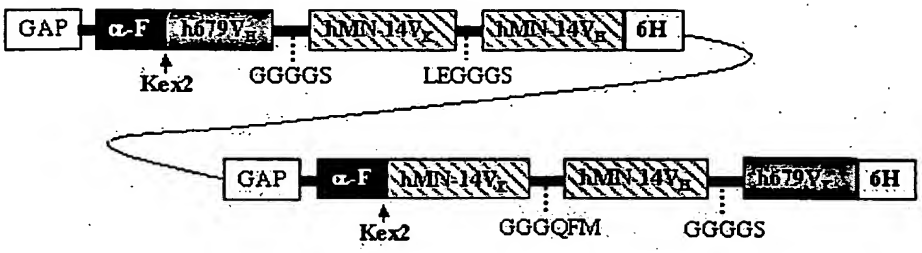
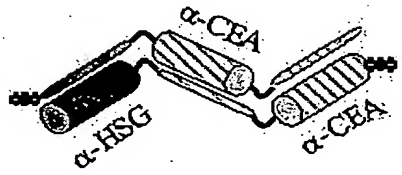
A.



B.

C.



BS14HP

Figure 1

FIGURE 1D

1) Amino acid sequence of Polypeptide 1. EAEAEFM-h679VH-GGGGS-hMN-14VK-LEGGGS-hMN-14VH-VD6His.

EAEAEFMEVQ LVESGGDLVK PGGSLKLSCA ASGFTFS**IYT MS**WLRQTPGK
CDR1h679VH

GLEWVA**TLSG DGDDIYYP**DS VKGRFTISRD NAKNSLYLQM NSLRAEDTAL
CDR2h679VH

YYCAR**VRLGD WDFDV**WGQGT TVSVSS**GGGG S**DIQLTQSPS SLSASVGDRV
CDR3h679VH linker

TITC**KASQDV GTSVA**WYQQK PGKAPKLLIY **WTSTRHT**GVP SRFSGSGSGT
CDR1hMN14VK CDR2hMN14VK

DFTFTISSLQ PEDIATYYC**Q QYSLYRS**FGQ GTKVEIKR**LE GGGS**EVQLVE
CDR3hMN14VK linker

SGGGVVQPGR SLRLSCSASG FDFT**TYWMS**W VRQAPGKGLE WIG**EIHPDSS**
CDR1hMN14VH CDR2hMN14VH

TINYAPSLKD RFTISRDNAK NTLFLQMDSL RPEDTGVYFC AS**LYFGFPWF**
CDR2hMN14VH CDR3hMN14VH

AYWGQGTPVTVS**VDHHHHHH**
CDR3hMN14VH 6His

Nucleic acid sequence (cDNA) of BS14HP polypeptide 1

GAGGCTGAAG CTGAATTCAT GGAAGTGCAG CTGGTGGAGT CAGGGGGAGA
CTTAGTGAAG CCTGGAGGGT CCCTGAAACT CTCCTGTGCA GCCTCTGGAT
TCACTTTCAG TATTTACACC ATGTCTTGGC TTCGCCAGAC TCCGGGAAAG
GGGCTGGAGT GGGTCGCAAC CCTGAGTGGT GATGGTGATG ACATCTACTA
TCCAGACAGT GTGAAGGGTC GATTCACCAT CTCCAGAGAC AATGCCAAGA
ACAGCCTATA TCTGCAGATG AACAGTCTAA GGGCTGAGGA CACGGCCTTG
TATTACTGTG CAAGGGTGCG ACTTGGGGAC TGGGACTTCG ATGTCTGGGG
CCAAGGGACC ACGGTCTCCG TCTCCTCAGG AGGTGGCGGA TCCGACATCC
AGCTGACCCA GAGCCCAAGC AGCCTGAGCG CCAGCGTGGG TGACAGAGTG
ACCATCACCT GTAAGGCCAG TCAGGATGTG GGTACTTCTG TAGCTTGGTA
CCAGCAGAAG CCAGGTAAGG CTCCAAAGCT GCTGATCTAC TGGACATCCA
CCCGGCACAC TGGTGTGCCA AGCAGATTCA GCGGTAGCGG TAGCGGTACC
GACTTCACCT TCACCATCAG CAGCCTCCAG CCAGAGGACA TCGCCACCTA
CTACTGCCAG CAATATAGCC TCTATCGGTC GTTCGGCCAA GGGACCAAGG
TGGAAATCAA ACGTCTCGAG GGCGGAGGTA GCGAGGTCCA ACTGGTGGAG
AGCGGTGGAG GTGTTGTGCA ACCTGGCCGG TCCCTGCGCC TGTCCTGCTC
CGCATCTGGC TTCGATTTCA CCACATATTG GATGAGTTGG GTGAGACAGG
CACCTGGAAA AGGTCTTGAG TGGATTGGAG AAATTCATCC AGATAGCAGT

ACGATTAACT ATGCGCCGTC TCTAAAGGAT AGATTTACAA TATCGCGAGA
CAACGCCAAG AACACATTGT TCCTGCAAAT GGACAGCCTG AGACCCGAAG
ACACCGGGGT CTATTTTTGT GCAAGCCTTT ACTTCGGCTT CCCCTGGTTT
GCTTATTGGG GCCAAGGGAC CCCGGTCACC GTCTCCGTCG ACCATCATCA
TCATCATCAT

FIGURE 1E

3) Amino acid sequence of polypeptide 2. EAEAEF-hMN-14VK-GGGQFM-hMN-14VH-GGGGS-h679VK-LD6His.

EAEAEFDIQL TQSPSSLSAS VGDRVTITC**K ASQDVGTSVA** WYQQKPGKAP
CDR1hMN14VK

KLLIY**WTSTR HT**GVPSRFSG SGSGTDFTFT ISSLQPEDIA TYYC**QQYSLY**
CDR2hMN14VK CDR3hMN14VK

RSFGQGTKVE IKR**GGGQFM**E VQLVESGGGV VQPGRSLRLS CSASGFDFT**T**
CDR3hMN14VK linker CDR1hMN14VH

YWMSWVRQAP GKGLEWIG**EI HPDSSTINYA PSLKD**RFTIS RDNAKNTLFL
CDR1hMN14VH CDR2hMN14VH

QMDSLRPEDT GVYFCAS**LYF GFPWFAY**WGQ GTPVTVS**GGG GS**DIVMTQSP
CDR3hMN14VH linker

SSLAVSPGER VTLTC**KSSQS LFNSRTRKNY LG**WYQQKPGQ SPKLLIY**WAST**
CDR1h679VK CDR2h679VK

RESGVPDRFS GSGSGTDFTL TINSLQAEDV AVYYC**TQVYY LCT**FGAGTKLE
CDR2h679VK CDR3h679VK

LKRLDHHHHH H
6His

Nucleic acid sequence (cDNA) of BS14HP polypeptide 2

GAGGCTGAAG CTGAATTCGA CATCCAGCTG ACCCAGAGCC CAAGCAGCCT
GAGCGCCAGC GTGGGTGACA GAGTGACCAT CACCTGTAAG GCCAGTCAGG
ATGTGGGTAC TTCTGTAGCT TGGTACCAGC AGAAGCCAGG TAAGGCTCCA
AAGCTGCTGA TCTACTGGAC ATCCACCCGG CACACTGGTG TGCCAAGCAG
ATTCAGCGGT AGCGGTAGCG GTACCGACTT CACCTTCACC ATCAGCAGCC
TCCAGCCAGA GGACATCGCC ACCTACTACT GCCAGCAATA TAGCCTCTAT
CGGTCGTTCG GCCAAGGGAC CAAGGTGGAA ATCAAACGTG GAGGTGGCCA
ATTCATGGAG GTCCAACTGG TGGAGAGCGG TGGAGGTGTT GTGCAACCTG
GCCGGTCCCT GCGCCTGTCC TGCTCCGCAT CTGGCTTCGA TTTCACCACA
TATTGGATGA GTTGGGTGAG ACAGGCACCT GGAAAAGGTC TTGAGTGGAT
TGGAGAAATT CATCCAGATA GCAGTACGAT TAACTATGCG CCGTCTCTAA
AGGATAGATT TACAATATCG CGAGACAACG CCAAGAACAC ATTGTTCCTG
CAAATGGACA GCCTGAGACC CGAAGACACC GGGGTCTATT TTTGTGCAAG
CCTTTACTTC GGCTTCCCCT GGTTTGCTTA TTGGGGCCAA GGGACCCCGG
TCACCGTCTC CGGAGGCGGT GGATCCGACA TTGTGATGAC ACAATCTCCA

TCCTCCCTGG CTGTGTCACC CGGGGAGAGG GTCACTCTGA CCTGCAAATC
CAGTCAGAGT CTGTTCAACA GTAGAACCCG AAAGAACTAC TTGGGTTGGT
ACCAGCAGAA ACCAGGGCAG TCTCCTAAAC TTCTGATCTA CTGGGCATCT
ACTCGGGAAT CTGGGGTCCC TGATCGCTTC TCAGGCAGTG GATCCGGAAC
AGATTTCACT CTCACCATCA ACAGTCTGCA GGCTGAAGAC GTGGCAGTTT
ATTACTGCAC TCAAGTTTAT TATCTGTGCA CGTTCGGTGC TGGGACCAAG
CTGGAGCTGA AACGGCTCGA CCATCATCAT CATCATCAT

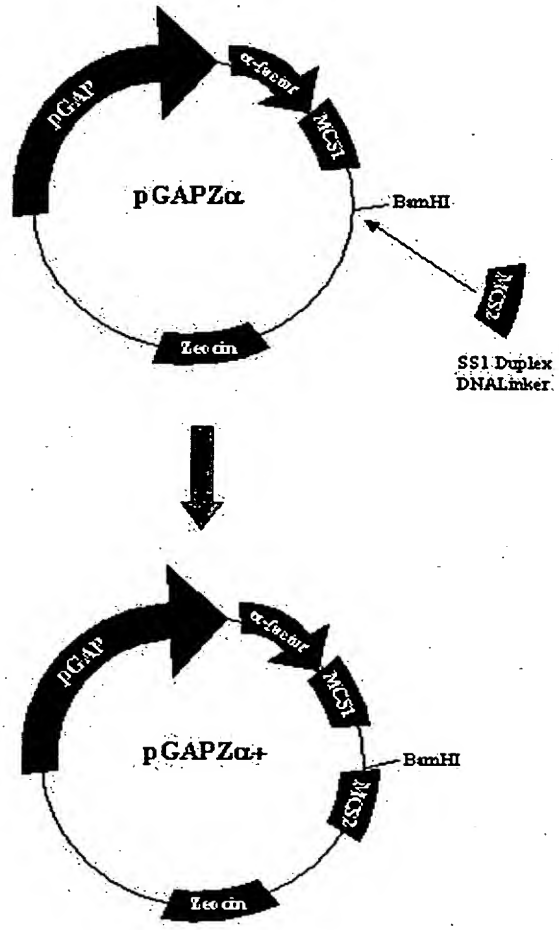


Figure 2

BIAcore analysis of BS14HP

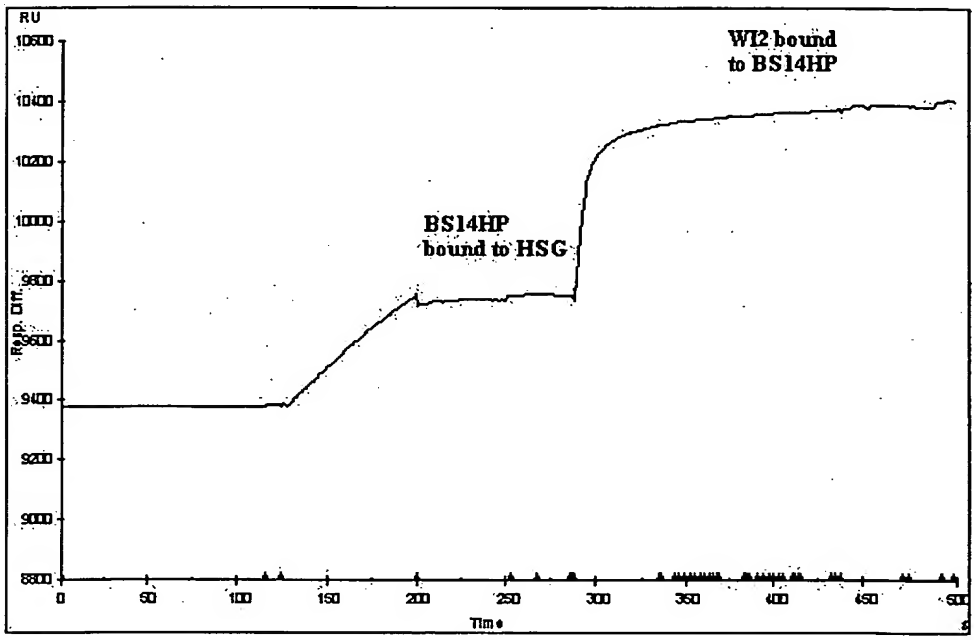
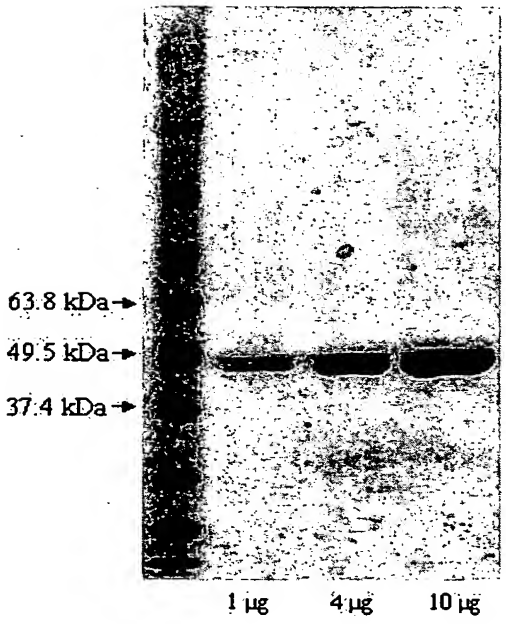


Figure 3

SDS-PAGE Analysis of BS14HP
B/N 111802



Calculated MW of polypeptides
40.06 kDa & 40.61 kDa

Figure 4

Size exclusion HPLC analysis

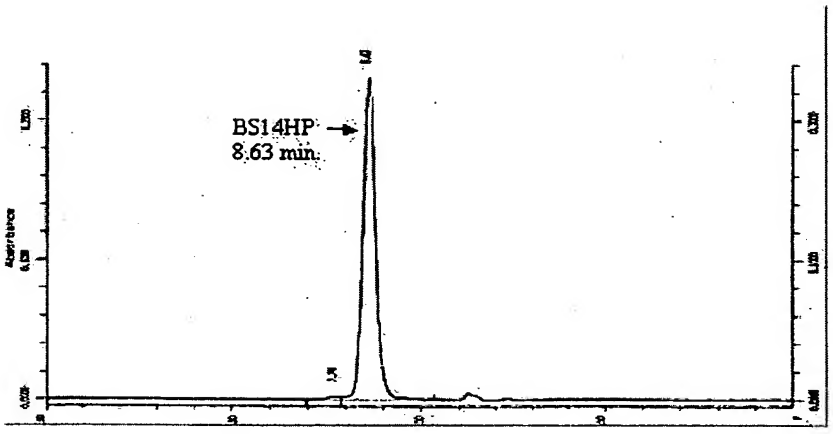
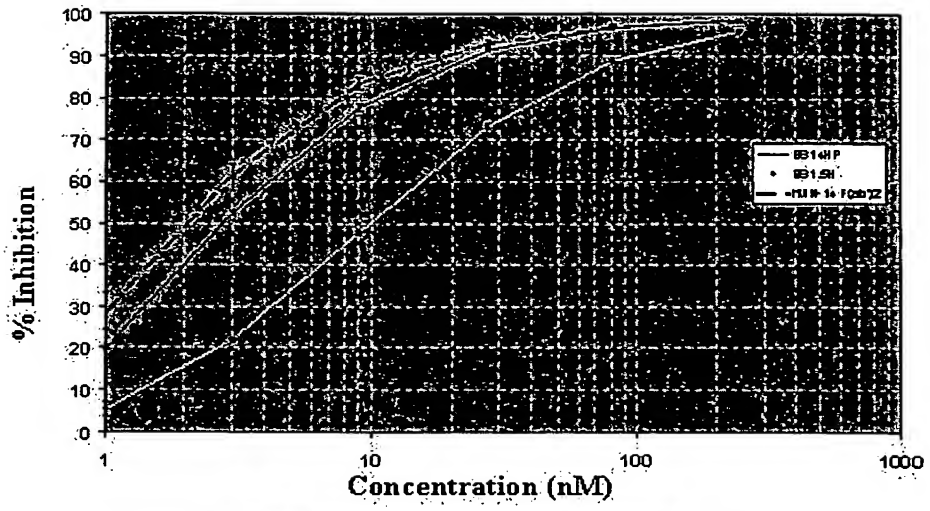


Figure 5

Competitive ELISA assay for CEA binding



	IC_{50}
BS14HP	2.7 nM
hMN-14 F(ab')2	2.0 nM
BS1.5H	10.0 nM

Figure 6

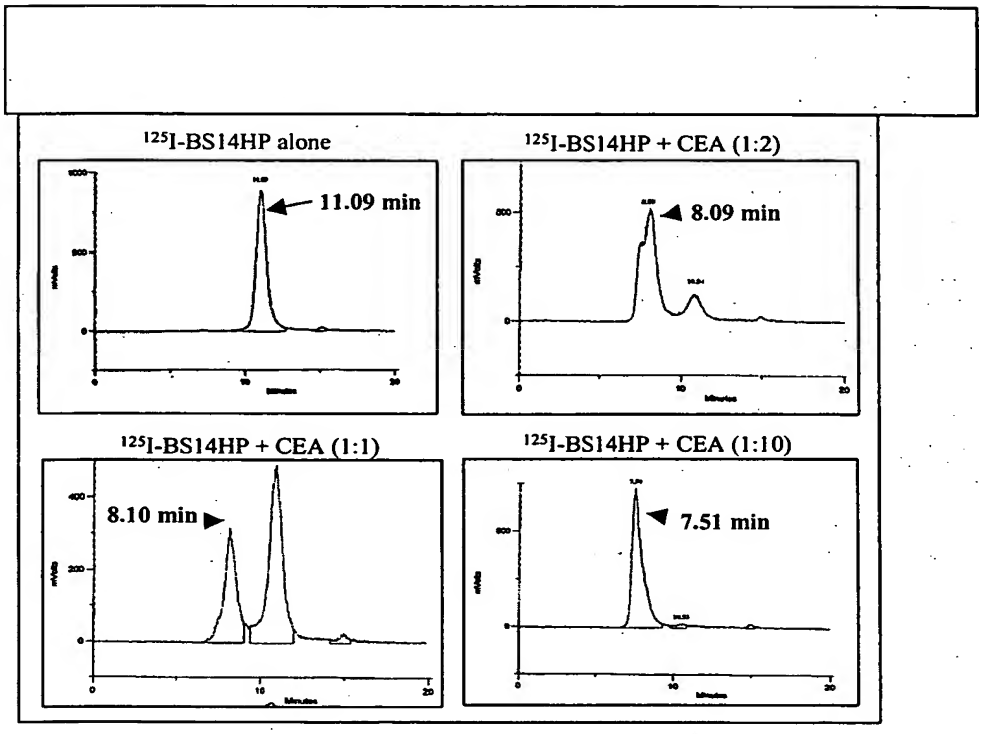


Figure 6B SE-HPLC Analysis of BS14HP immunoreactivity with CEA

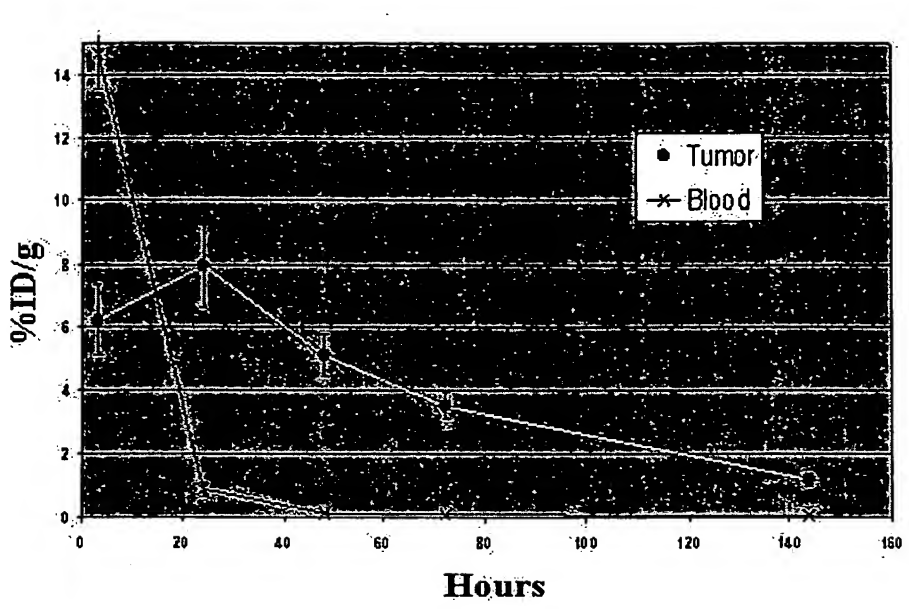
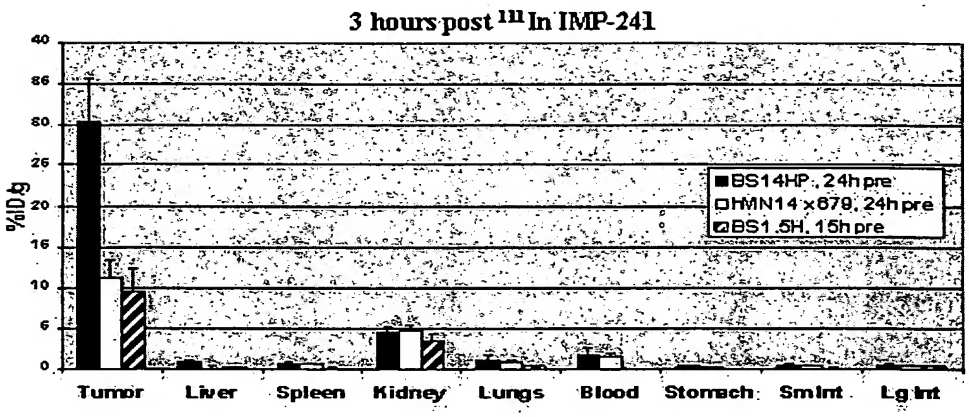


Figure 7

A. Biodistribution of ^{111}In-IMP241in mice pretargeted with BS14HP, BS1.5H or hMN14 x 679 Fab' x Fab'



B. Tumor/non-tumor ratios of ^{111}In-IMP241in mice pretargeted with BS14HP, BS1.5H or hMN-14 x 679 Fab' x Fab'

	BS14HP	hMN-14 x 679	BS1.5H
Liver	36.19 (±18.8)	22.20 (±6.3)	120.00 (±36.0)
Spleen	57.39 (±46.0)	27.80 (±5.9)	181.00 (±58.0)
Kidney	6.70 (±0.7)	2.50 (±0.5)	2.98 (1.1±)
Lungs	29.94 (±15.6)	14.10 (±2.8)	48.60 (±19.3)
Blood	20.32 (±34.7)	8.10 (±2.1)	284.00 (±50.6)
Stomach	123.12 (±242.0)	103.00 (±15.2)	530.00 (±291.7)
Sm. Intestine	78.95 (±55.0)	53.40(±14.4)	235.00 (±138.7)
Lg. Intestine	80.94 (±26.7)	37.40 (±9.2)	61.20 (±33.2)

Figure 8

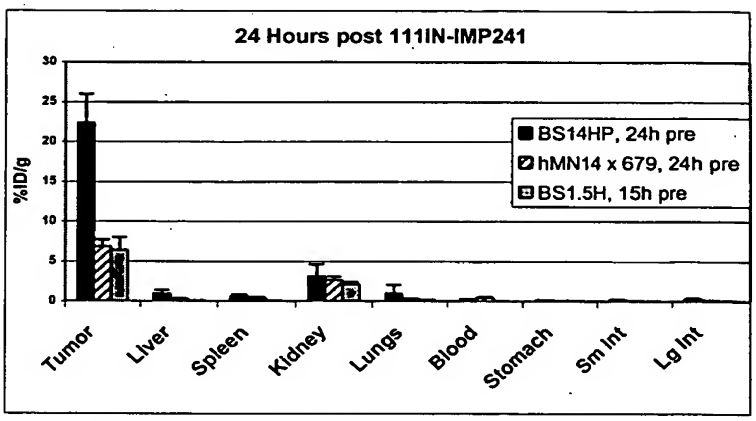

24 Hours post 111IN-IMP241
%ID/g
30
25
20
15
10
5
0
BS14HP, 24h pre
hMN14 x 679, 24h pre
BS1.5H, 15h pre
Tumor
Liver
Spleen
Kidney
Lungs
Blood
Stomach
Sm Int
Lg Int

Figure 8C

IMP 281

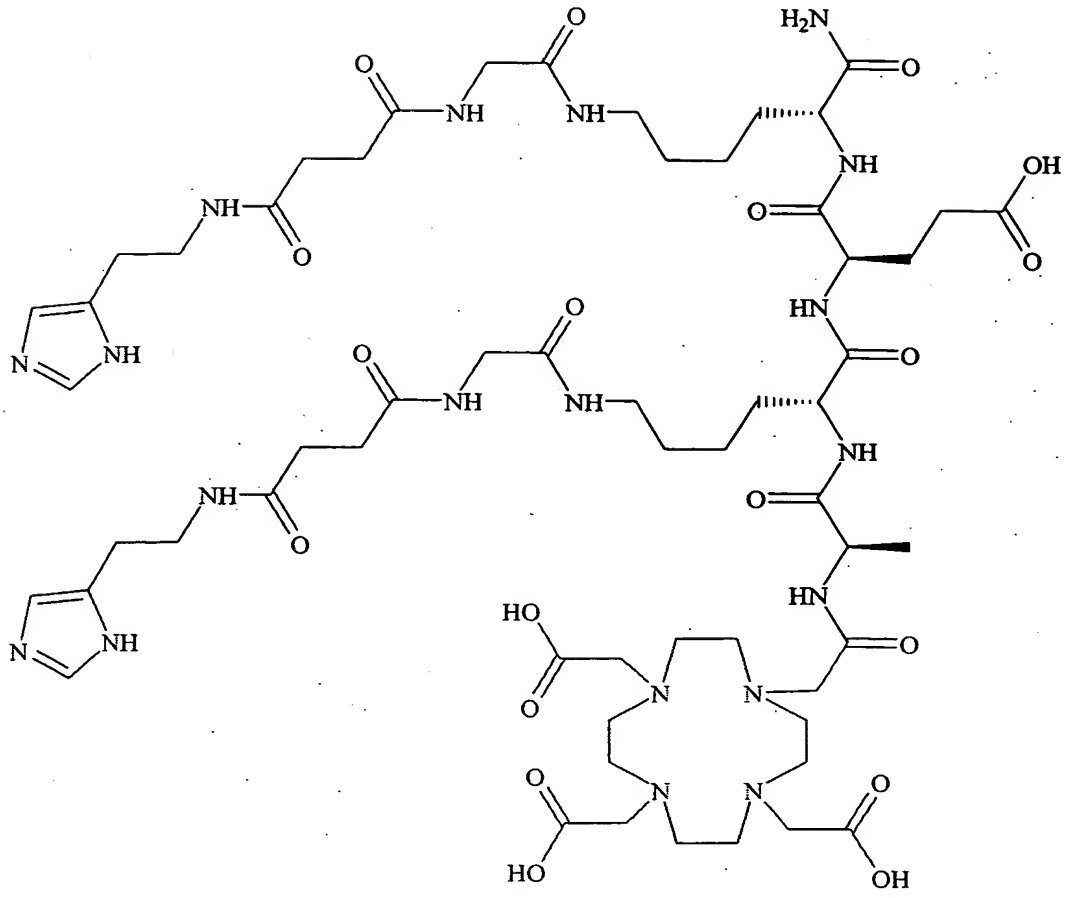


Figure 9A

IMP 284

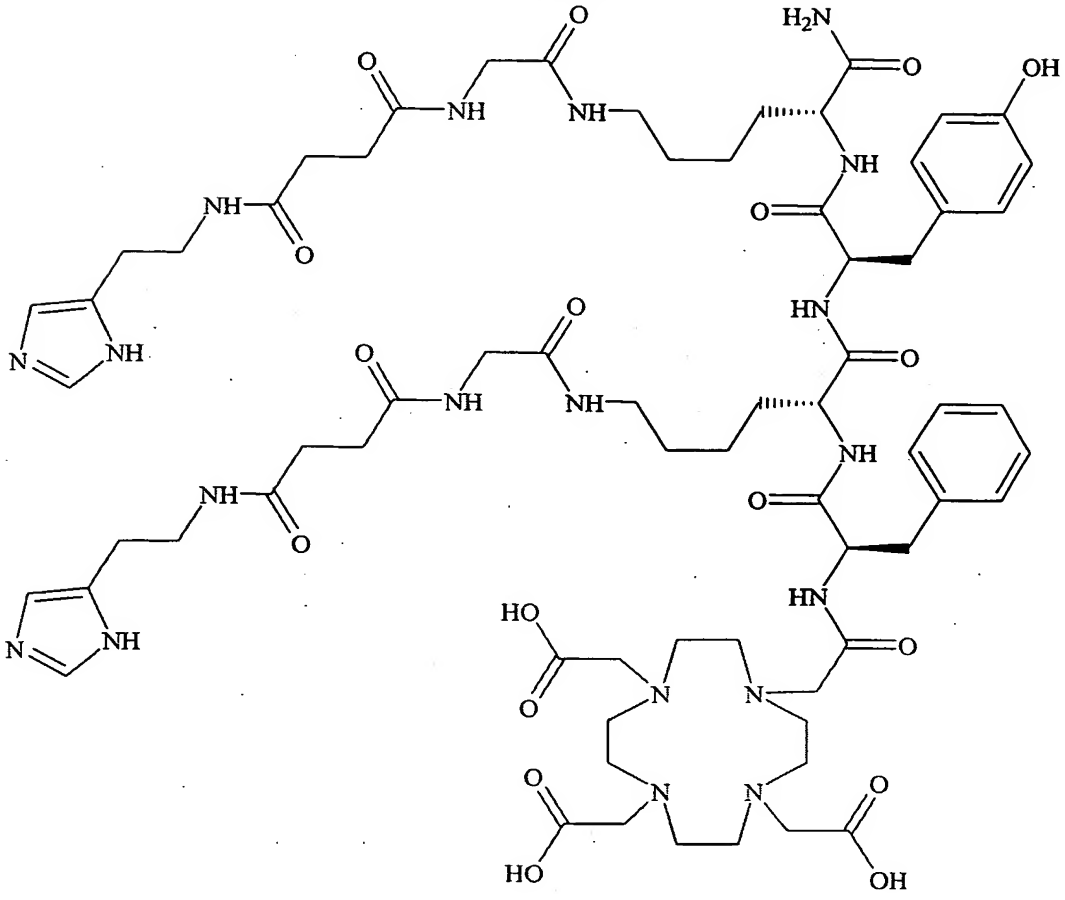


Figure 9B

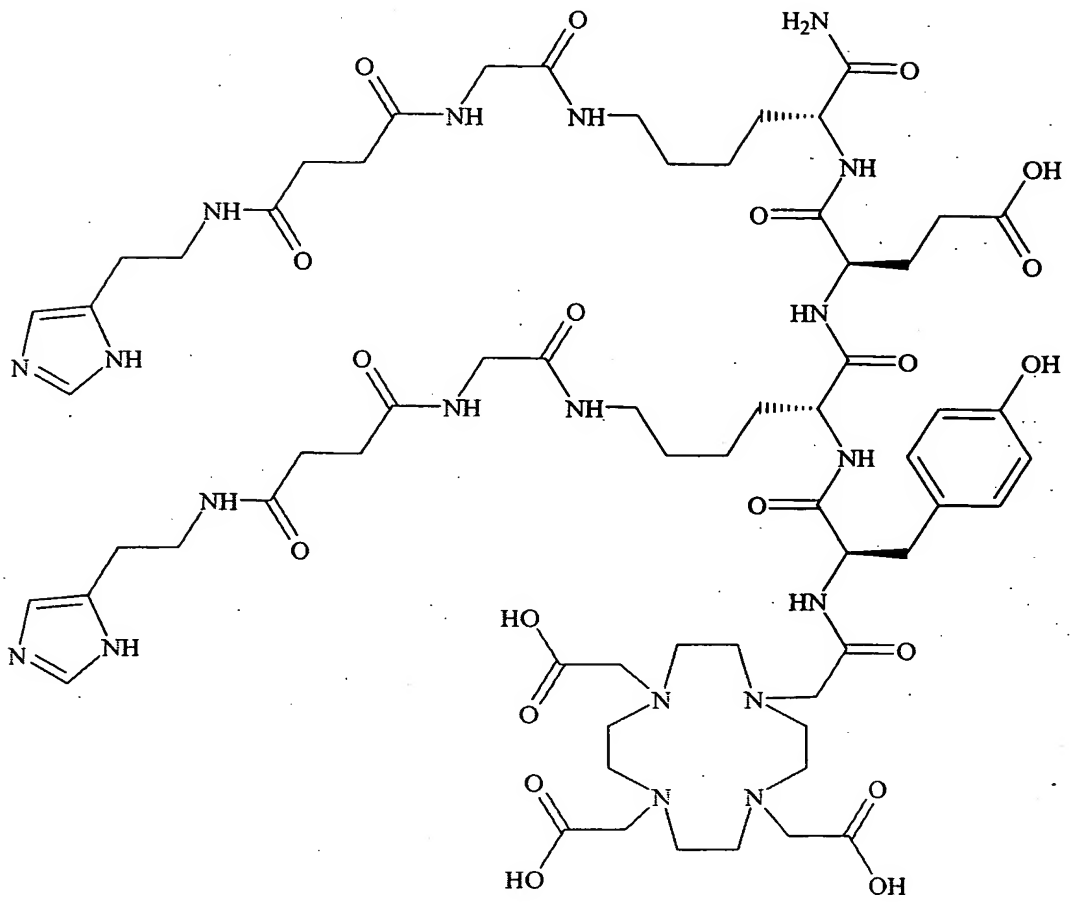


Figure 9C

Functional features of the SV3 shuttle vector.

HindIII—XhoI—XbaI—Leader peptide—NcoI—SalI—6His—Stop—Stop—BglII—EagI—EcoRI

Figure 10 A

Features of the ORF/Polypeptide 1, and ORF/Polypeptide 2

ORF1/Polypeptide 1
Ldr Pep—h679V_H—GGGGS—hMN-14V_K—LEGGGS—hMN-14V_H—HHHHHH

ORF2/Polypeptide 2
Ldr Pep—hMN-14V_K—GGGQFM—hMN-14V_H—GGGGS— h679V_k—HHHHHH

Figure 10B

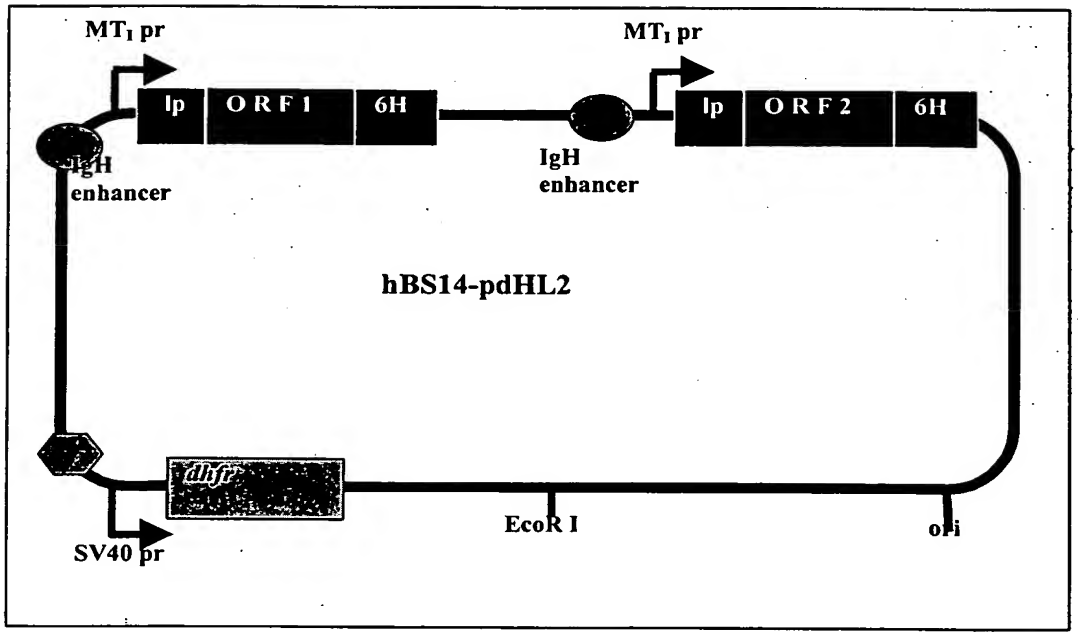


Figure 11. Schematic representation of hBS14-pDHL2 expression vector.

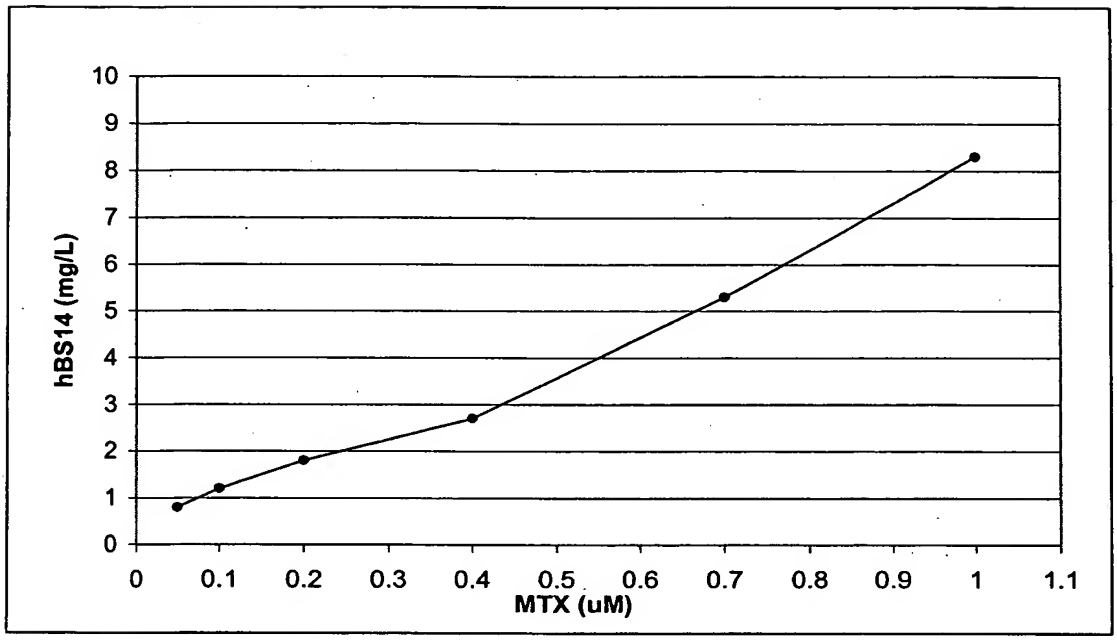
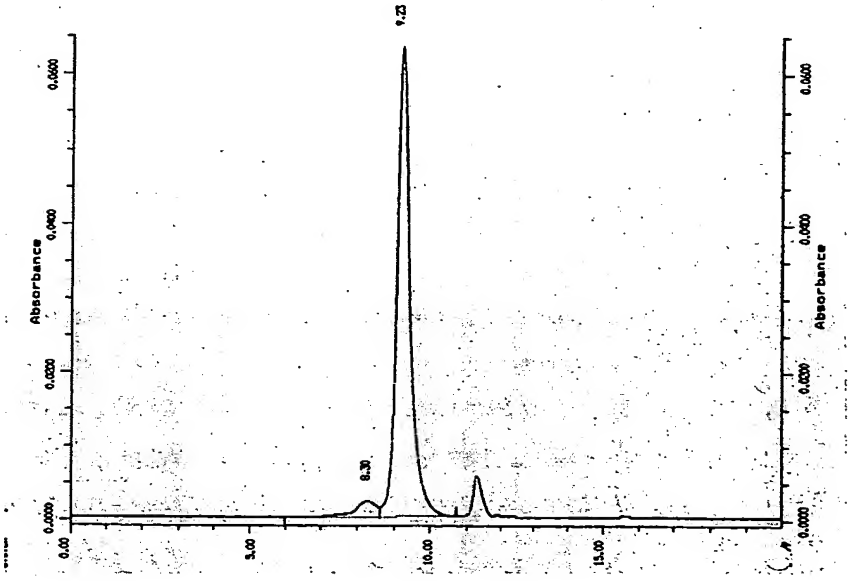


Figure 12. MTX amplification of hBS14 SP2/0 clone 1H6

hBS14 102003
YB2/0



hBS14 100103
SP2/0

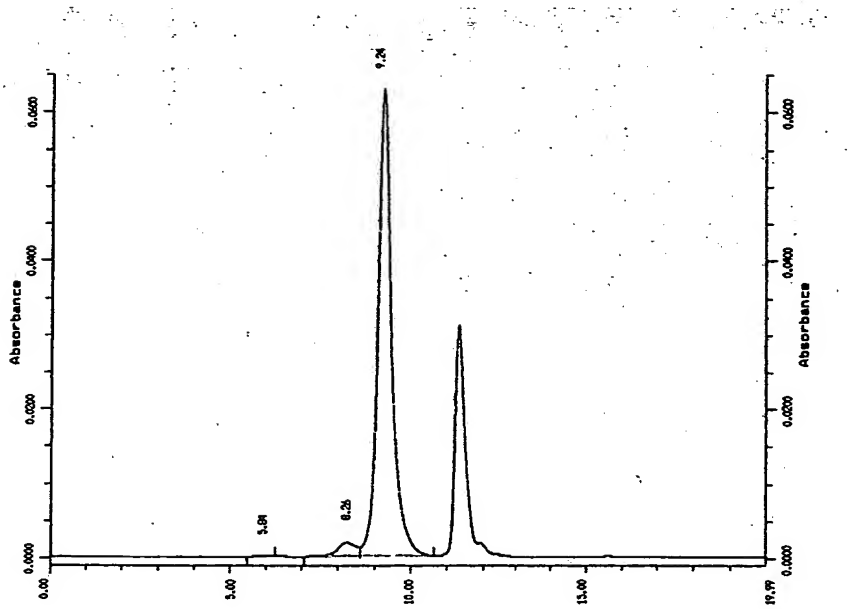


Figure 13. SE-HPLC analysis of purified hBS14

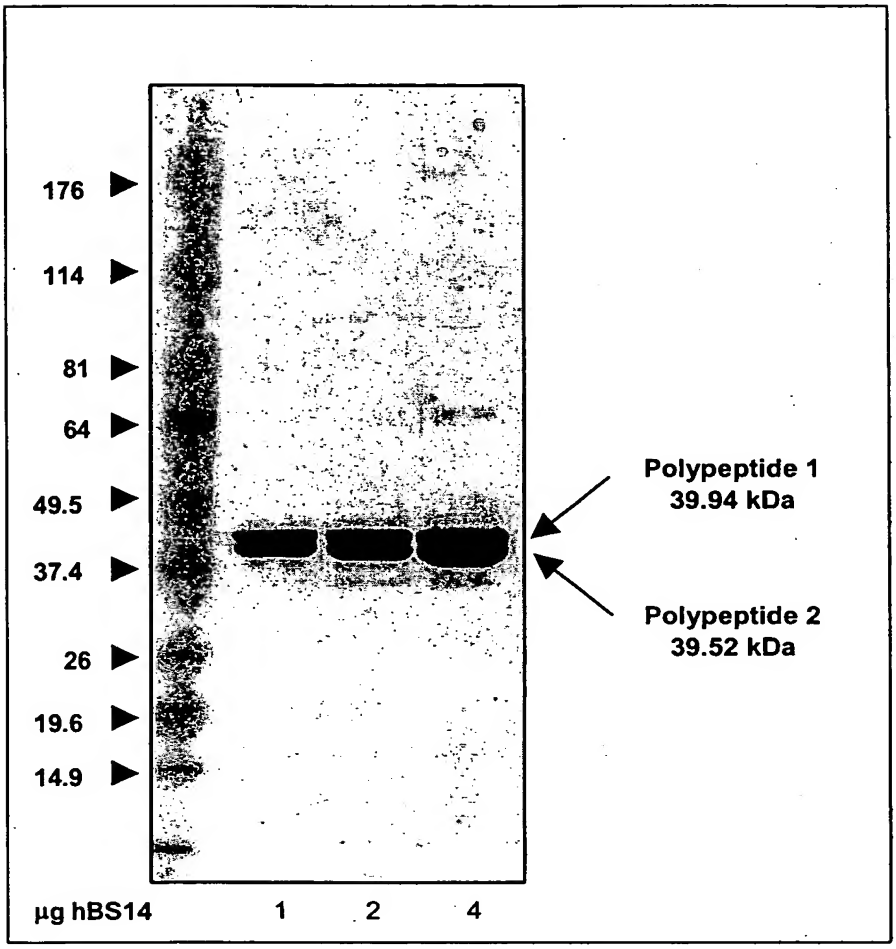


Figure 14. SDS-PAGE analysis of purified hBS14

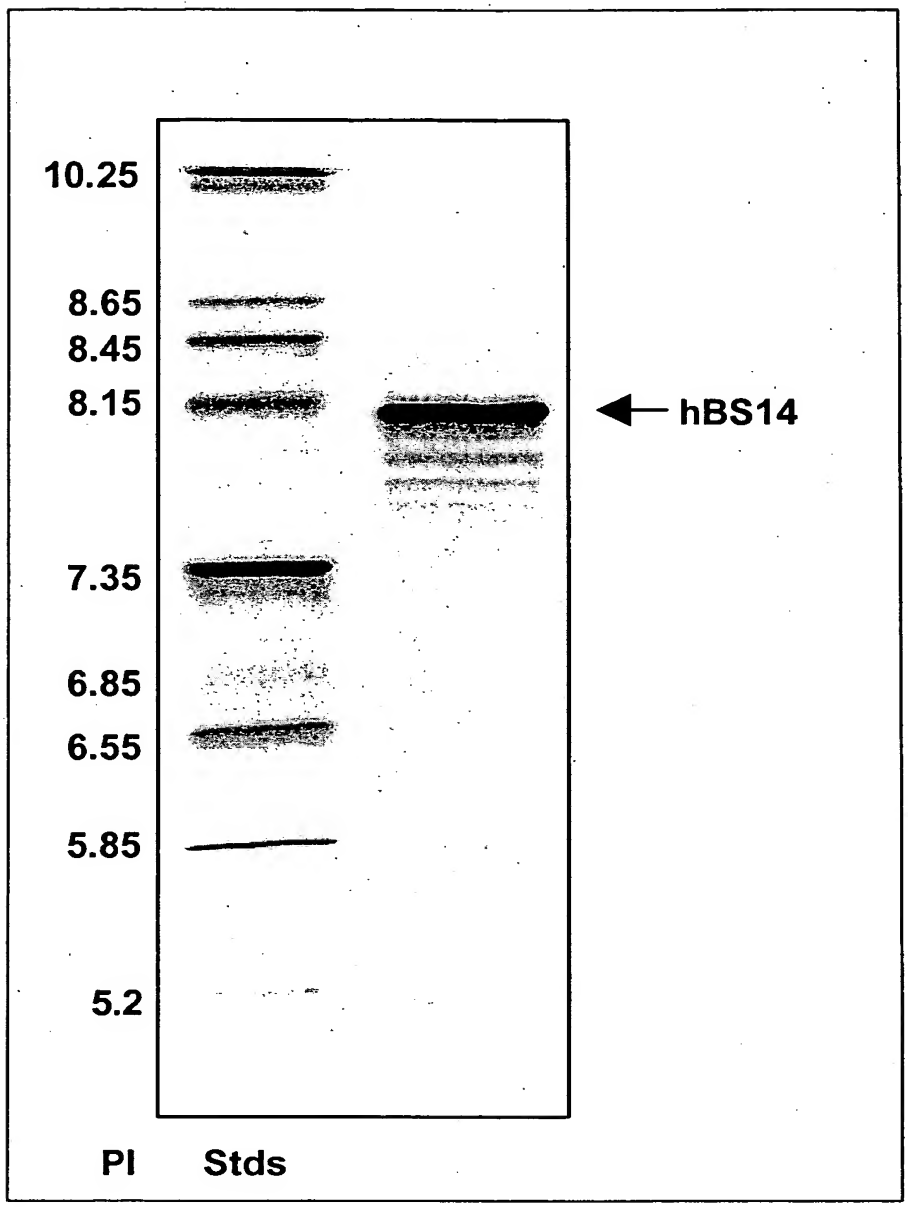


Figure 15. IEF analysis of purified hBS14

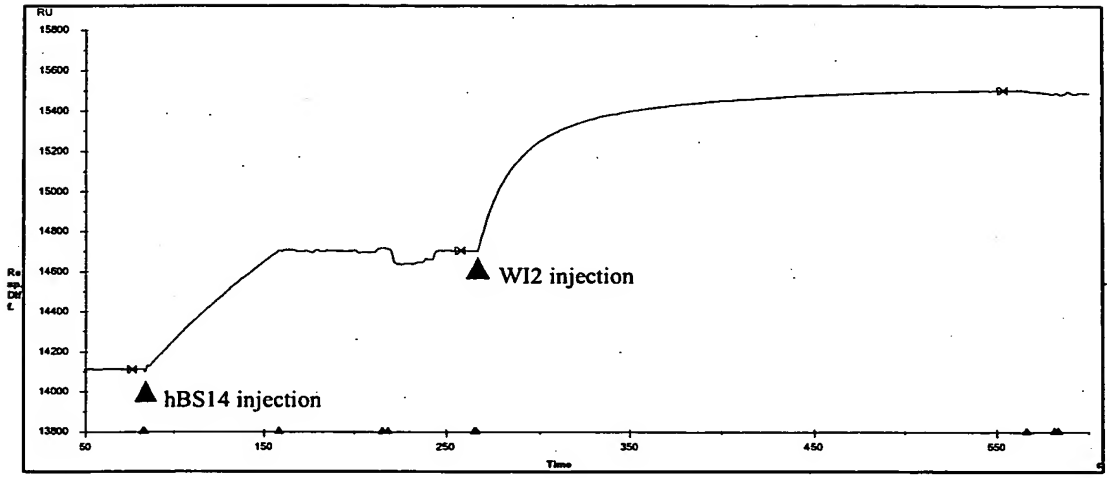


Figure 16. BIAcore analysis of hBS14

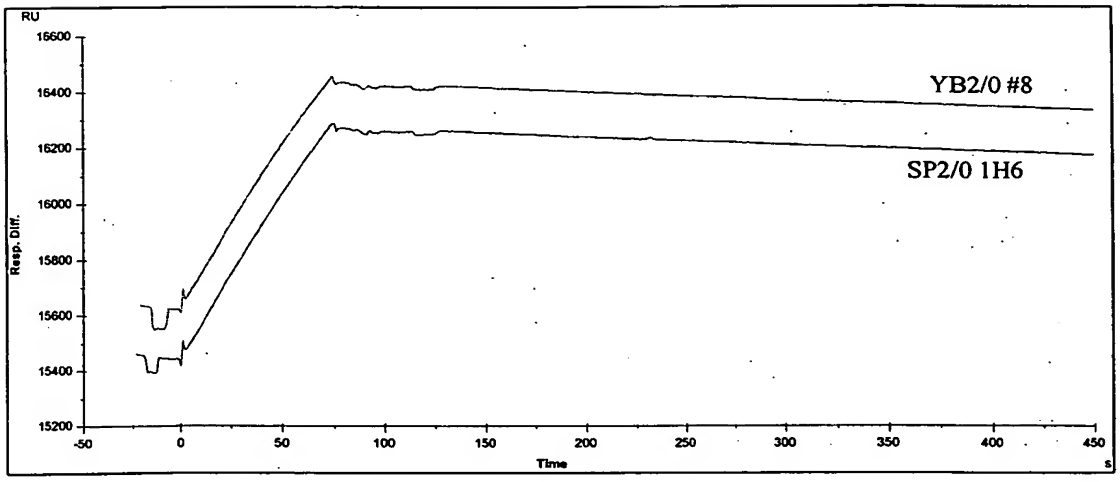


Figure 17. BIAcore analysis of HSG binding of hBS14 produced in either SP2/0 or YB2/0 cells

Figure 18

IMP 291 Ac-Lys(HSG-iAsp-)-Cys-NH_2 MH^+ 656

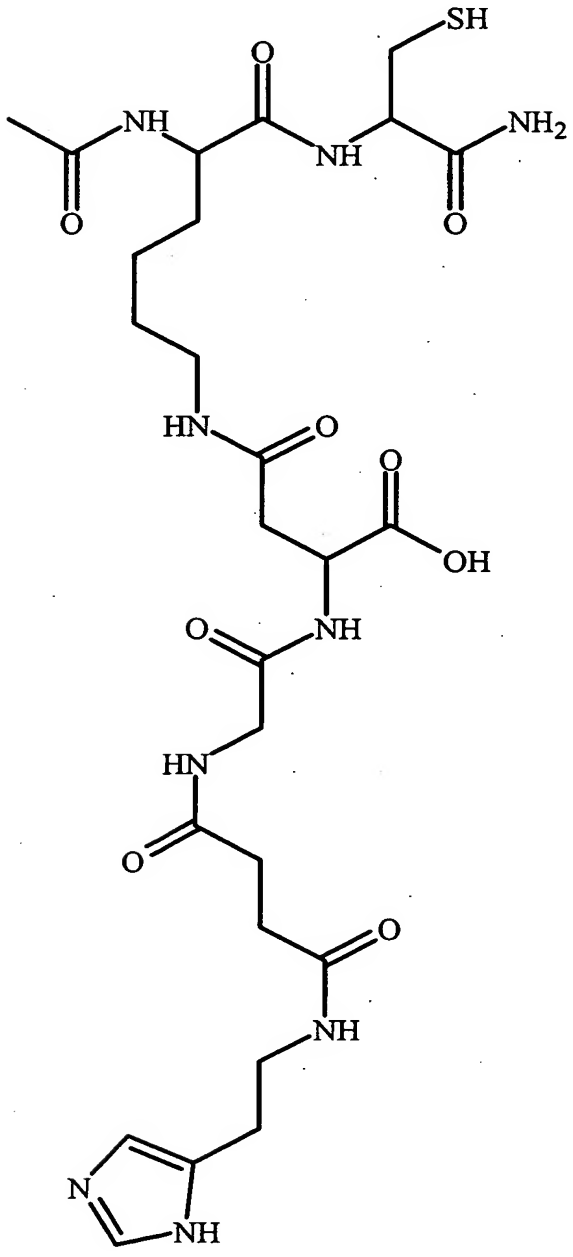


Figure 19

IMP 245 DOTA-Phe-Lys(HSG)-D-Tyr-Lys(HSG)-Lys(Tscg-Cys-)-NH_2 MH^+ 1832

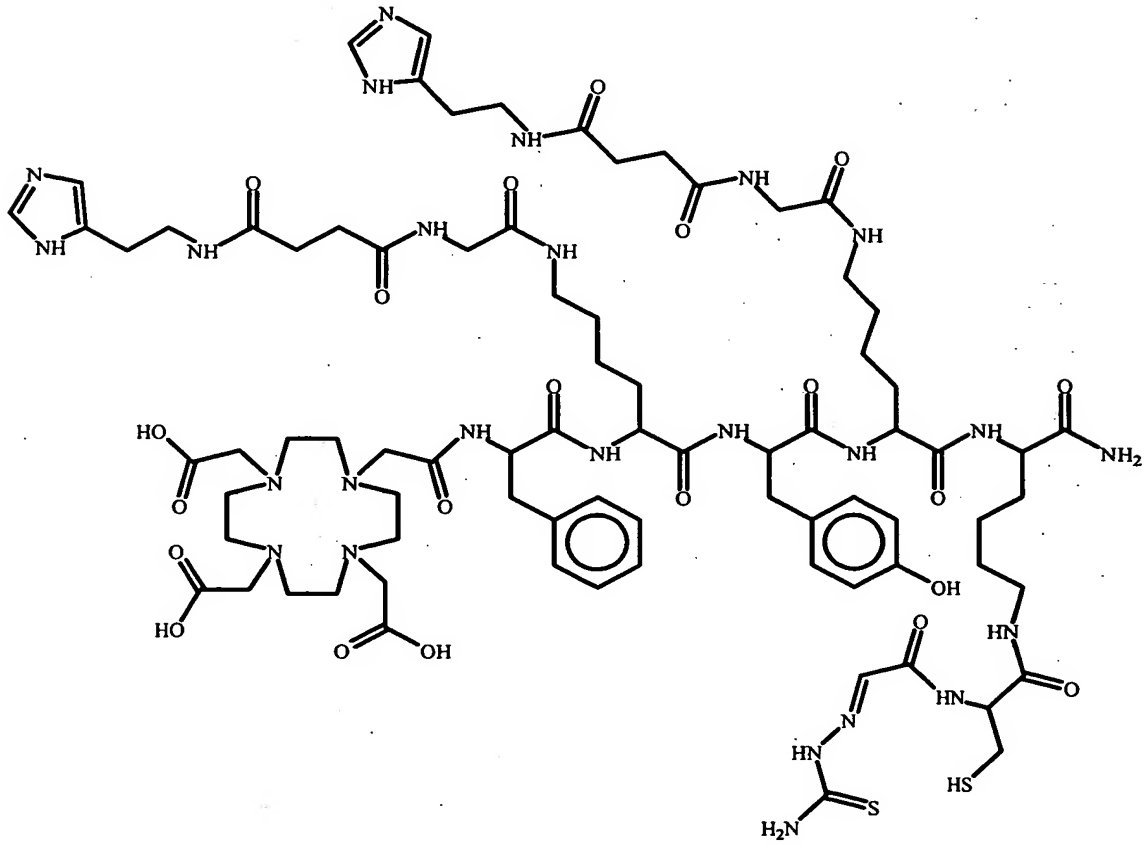
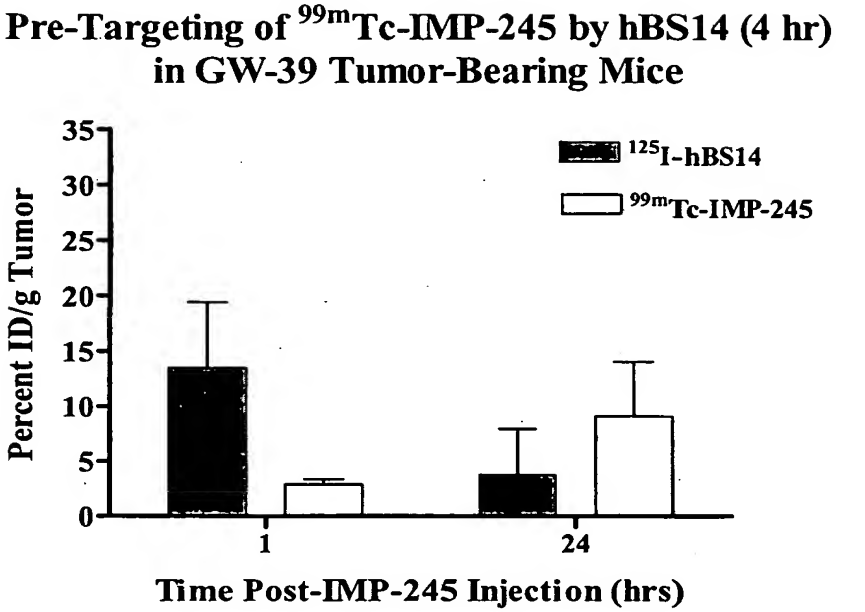
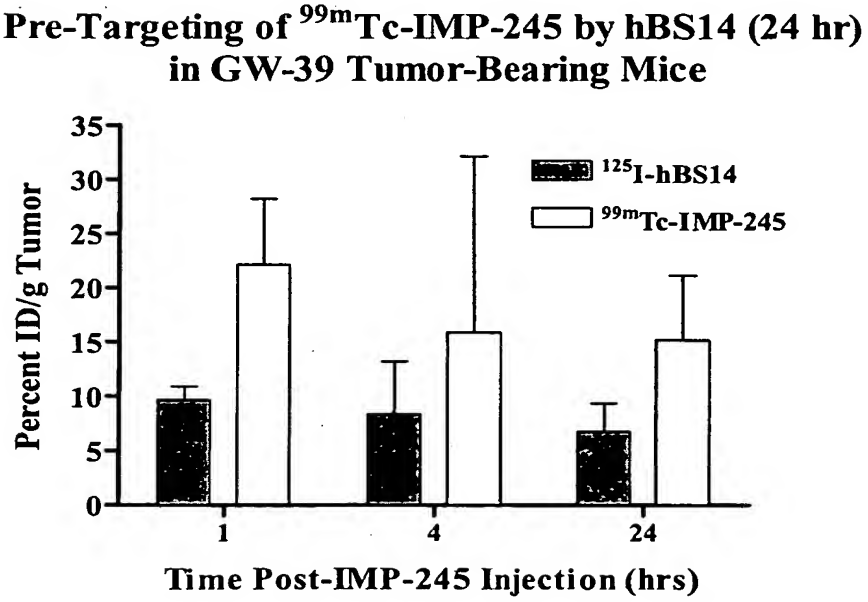


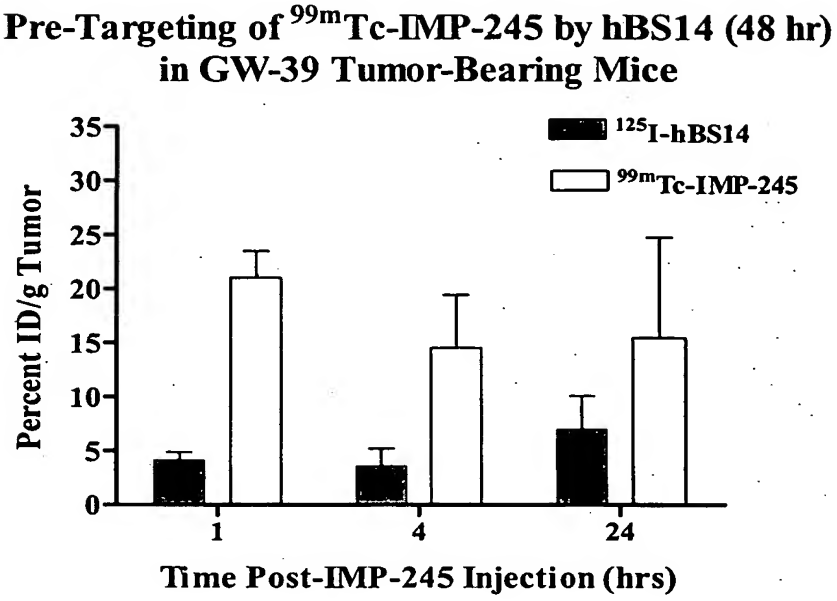
Figure 20


Pre-Targeting of 99mTc-IMP-245 by hBS14 (4 hr)
in GW-39 Tumor-Bearing Mice
125I-hBS14
99mTc-IMP-245
Percent ID/g Tumor
35
30
25
20
15
10
5
0
1
24
Time Post-IMP-245 Injection (hrs)


Pre-Targeting of 99mTc-IMP-245 by hBS14 (24 hr)
in GW-39 Tumor-Bearing Mice
125I-hBS14
99mTc-IMP-245
Percent ID/g Tumor
35
30
25
20
15
10
5
0
1
4
24
Time Post-IMP-245 Injection (hrs)

Edmund A. ROSSI et al.
"Polyvalent Protein Complex"
Attorney Docket No. 41133-0006US1

Figure 21



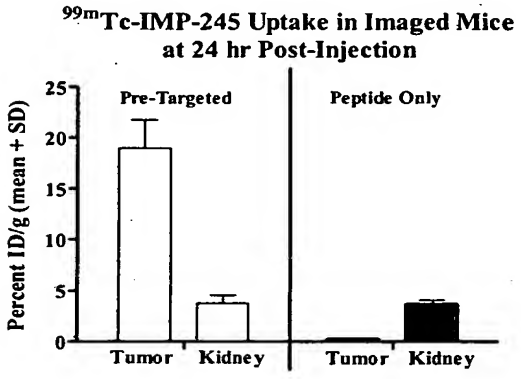


Figure 22

Percent ID/g and Tumor Non-Tumor Ratios of ^{99m}Tc-IMP-245 at 1 hr Post-Injection.

	4 hrs hBS14 Clearance		24 hrs hBS14 Clearance		48 hr hBS14 Clearance	
Tissue	%ID/g ± (SD)	T:NT Ratio ± (SD)	%ID/g ± (SD)	T:NT Ratio ± (SD)	%ID/g ± (SD)	T:NT Ratio ± (SD)
GW-39	2.9 ± 0.5	--	22.2 ± 6.1	--	21.0 ± 2.5	--
Liver	8.3 ± 0.6	0.4 ± 0.07	1.5 ± 0.8	17.5 ± 6.5	1.1 ± 0.2	19.7 ± 3.6
Spleen	7.5 ± 1.9	0.4 ± 0.08	0.9 ± 0.3	27.7 ± 9.4	0.5 ± 0.1	41.3 ± 9.7
Kidney	13.4 ± 1.1	0.2 ± 0.03	5.3 ± 0.8	4.1 ± 0.7	7.9 ± 0.8	2.7 ± 0.5
Lungs	10.7 ± 3.1	0.3 ± 0.10	1.7 ± 0.7	14.1 ± 4.8	1.0 ± 0.4	23.0 ± 6.8
Blood	36.9 ± 6.4	0.1 ± 0.01	7.1 ± 8.7	5.7 ± 3.4	1.6 ± 0.3	13.6 ± 2.3
Stomach	1.3 ± 0.4	2.4 ± 0.67	4.0 ± 7.6	29.4 ± 18.8	3.5 ± 0.9	6.4 ± 2.0
Small Int.	3.7 ± 0.3	0.8 ± 0.07	3.9 ± 5.3	11.8 ± 6.4	2.4 ± 0.5	8.8 ± 1.5
Large Int.	2.2 ± 1.7	1.8 ± 0.98	0.4 ± 0.2	69.4 ± 31.2	0.4 ± 0.1	56.0 ± 11.5
Muscle	1.2 ± 0.2	2.4 ± 0.32	4.8 ± 6.4	19.1 ± 26.3	4.1 ± 5.0	18.4 ± 25.6
Tumor Weight (grams)	0.309 ± 0.139		0.309 ± 0.136		0.972 ± 0.640	

Edmund A. ROSSI et al.
"Polyvalent Protein Complex"
Attorney Docket No. 41133-0006US1

Figure 23